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DEVELOPMENT OF ANAMMOX PROCESS FOR ANIMAL WASTE TREATMENT: EXPERIENCES IN BRAZIL

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Abstract. The isolation of anaerobic ammonium oxidation (anammox) bacteria adapted to animal wastewater environments can be of significant importance to farming systems, because excess ammonia in modern, industrial-type livestock production is a global problem, and the use of conventional biological nitrogen (n) removal methods is usually hindered by cost; thus, we think that the more economical anammox based treatment can greatly facilitate adoption of advanced wastewater treatment technologies by farmers. research was conducted to develop process applications for anammox bacteria acclimated to animal wastewater conditions using microbial immobilization techniques. Traditionally, removal of N from wastewater uses a combination of nitrification and denitrification. Nitrification consumes large amounts of oxygen to convert NH_4^+ into nitrate (NO_3^-), while denitrification requires addition of organic carbon to convert NO_3^- into N_2 gas. The sludges containing anammox bacteria were obtained from a swine farm in Santa Catarina, Brazil. The sludges were obtained from sediment in an old (inactive) anaerobic lagoon used to treat swine manure. Laboratory bioreactors were seeded with the manure sludges after acclimation with nitrate solution to remove endogenous carbon. A distinct red biomass growth, which is typical of the anammox planctomycete bacteria, developed in the reactors. Under these conditions and protocol, it took about 75 days for the anammox reaction to develop from farm sludges. As biomass carrier, the reactors used a net type acryl-resin fiber material designed to enhance retention of microorganisms. These findings overall may lead to development of more economical treatment systems for livestock wastewater and other effluents containing high ammonia concentration.

Keywords. Animal manure, nitrogen removal and environmental impact.

Introduction

Brazil is the fourth world producer of swine meat with 32,938,000 heads and Santa Catarina State is the national leader in swine production with 6,006,000 heads (Anualpec, 2006). While this is a very important activity in the agribusiness, it has a significant environmental impact mainly in regions with high livestock concentration.

To reduce the pollution of the activity some initiatives need be held. Many treatment technologies have been successfully developed to remove organic load, but the nutrient (N and P) removal did not receive the same attention. This nutrient removal development needs to be addressed, mainly in areas that manure production excess the soil support capacity.

In the case of nitrogen in swine manure, it is present in a high concentration, in several forms and oxidation states. The most representative forms are organic nitrogen, ammonium/ammonia (NH_4^+/NH_3) and, during the treatment nitrite (NO_2^-) and nitrate (NO_3^-). In the conventional process to remove nitrogen (nitrification/denitrification), autotrophic and heterotrophic microorganisms grow in separate optimized reactors in aerobic and anaerobic conditions. This process involves high costs in implementation and operation (Toh et al., 2006).

Since the beginning of the last decade many new processes to remove nitrogen have been developed, among these the Anaerobic Ammonium Oxidation (anammox) has received special attention. In this process, ammonium (NH_4^+) is directly oxidized to N_2 with nitrite (NO_2^-) working as a electron acceptor. This process is autotrophic, using CO_2 as the carbon source. The global steichiometry of an anammox reactor according to Strous et al. (1998) is:

$$NH_4^+ + 1.32NO_2^- + 0.066HCO_3^- + 0.13H^+ \rightarrow 1.02N_2 + 0.26NO_3^- + 0.066CH_2O_{0,5}N_{0,15} + 2.03H_2O$$

In this work, the aim was to find anammox activity in a sample of sludge from swine manure treatment and increase the bacteria population by sludge acclimation and immobilization, feeding it with a synthetic medium to estimate the capacity to remove nitrogen.

Materials and Methods

Inocculum

The sludge for this study was collected from the bottom of an inactive anaerobic lagoon in experimental swine manure treatment lagoons at Embrapa's Swine and Poultry, Concordia, SC, Brazil.

Acclimation

The sludge was screened, washed with water, and kept in a KNO_3 (100 mg/L) solution until the denitrification process stopped, that is a good indication that the organic carbon was consumed. After this initial acclimation, the sludge was inoculated in an anammox reactor.

Reactor set-up

The sludge was inoculated at 4 g/L TSS in a 2 L reactor. The process was kept at 35 ^{0}C and pH 7. The feeding medium was prepared according to Shierholt Neto et al. (2006). To remove the interference of oxygen in the process, N_2 was bubbled until dissolved oxygen concentration was 0.5 mg/L. The Hydraulic Retention Time (HRT) was adjusted between 18.5 h and 24 h.

Chemical analyses

All chemical analyses were done according to APHA (1995). Liquid samples from the reactor (in and out) were taken twice a week and analyzed for N-NH_3, N-NO_2, N-NO_3 using Flow Injection Analysis (FIALAB INSTRUMENTS). Alkalinity was determined by titration with H_2SO_4 0.02 mol/L until pH 4.5.

Results and Discussion

The sludge acclimation lasted about two months, whereby organic carbon was being consumed with the added nitrate. At day 63, the denitrification process stopped. After this initial step, the reactor was inoculated and feed with the synthetic medium. It took around 75 days, after the KNO_3 addition procedure, until the immobilized sludge started to develop anammox activity (day zero in figure 1).

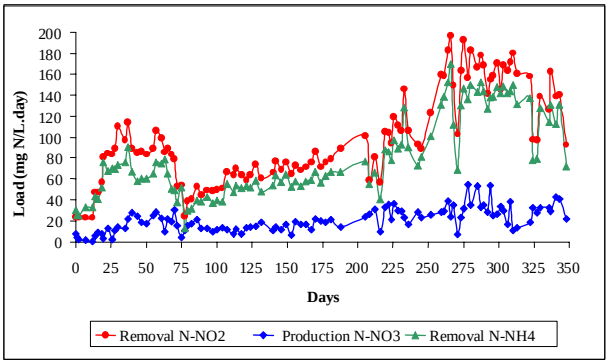

Figure 1. Nitrogen removal in the anammox reactor during the operation time.

In figure 1 it is possible to observe the capacity of the sludge to quickly start removing nitrogen, reaching around 200 mg/L.day approximately in 30 days. After this time the reactor almost collapsed because during a week the medium was prepared without carbonate alkalinity supplement. Therefore, alkalinity was not only important to buffer the pH, but also as a carbon source to the anammox bacteria. The reactor needed an additional 100 days to return to the same removal load. The reactor reached in day 266 the highest removal load, 350 mg/L.day with 85 % removal efficiency of nitrogen, showing the potential of the system to effluents with high concentration of nitrogen.

The reactor stoichiometric coefficients (Figure 2) were very close to those presented for anammox in the literature (Strous et al., 1998), showing that the acclimated swine manure sludge developed an anammox activity.

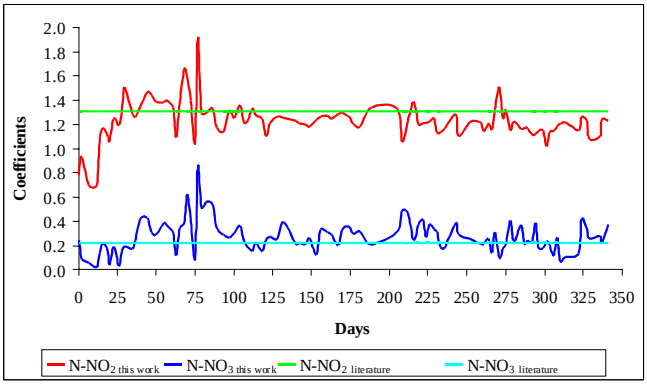

Figure 2. Stoichiometric coefficients of nitrite and nitrate related to ammonia, according to the reactor nitrogen mass balance.

Data in figure 3 show the correlation among NH_4^+:NO_2^-:NO_3^-. It is possible to observe a good correlation between ammonia and nitrite removal (r^2 = 0.97) and for N removed ($N\text{-}NH_4^+ + N\text{-}NO_2^-$) in the reactor, with the assumption that de nitrogen species are converted to N_2 (r^2 = 0.96).

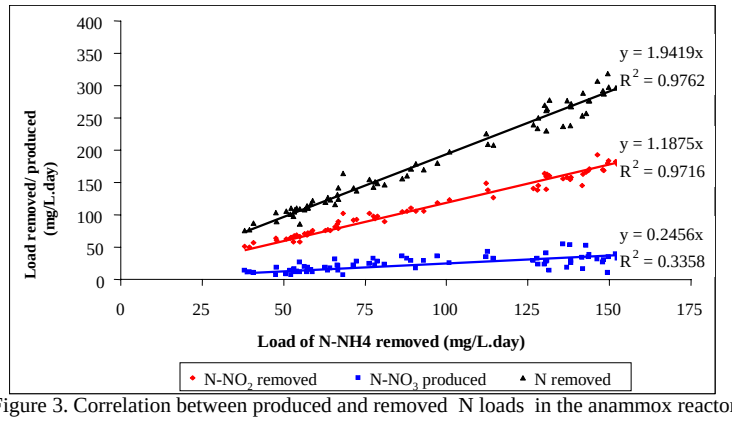

Figure 3. Correlation between produced and removed N loads in the anammox reactor.

The stoichiometric index for NH_4^+:NO_2^-:NO_3^- obtained was 1:1.18:0.24 that is similar to that found by Jetten et al., (1999) (1:1.31:0.22) in the Netherlands. and by Vanotti et al., 2006 (1:1.24:0.24) in the USA, confirming the anammox activity in the reactor in Brazil.

Conclusion

The sludge from swine manure treatment system was a good source to isolate and purify bacteria with anammox activity. This is important to develop a high-performing anammox culture that can grow in a high level of nitrogen concentration typical of animal wastewater. During the reactor operation time, the reactor reached a maximal removal nitrogen load of 0.35 Kg N/m^3 . day

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